11)-Fish granzymes and their role in the innate cell-madiated cytotoxicity against nodavirus-infected cells.

Elena Chaves-Pozo[1] , Yulema Valero[1] , María A. Esteban[2] , José Meseguer[2] , Alberto Cuesta[2]

1) Centro Oceanográfico de Murcia, Instituto Español de Oceanografía, Puerto de Mazarrón, 30860. Spain

2) Fish Innate Immune System Group, Department of Cell Biology and Histology, Faculty of Biology, Regional Campus of International Excellence "Campus Mare Nostrum", University of Murcia, 30100 Murcia, Spain

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Abstracts:

Granzymes (Gzm) are granule-associated serine proteases, which are important effector molecules in cell-mediated cytotoxicity, classified by its proteolytic activity in: tryptase (GzmA and GzmK), Asp-ase (GzmB), Met-ase (GzmM) or chymase (others). In fish, even at gene level, very few studies have partially identified and characterize them. In this work we have identified the activity of these four granzymes, using colorimetric assays, in leucocytes from gilthead seabream (*Sparus aurata*) and European sea bass (*Dicentrarchus labrax*), and how they are increased upon cytotoxic activity against nodavirus-infected tumor cells. The results also showed that GzmA was the most abundant and GzmB the lowest. Deeper characterization of the GzmB led us to identify that seabream leucocytes under cytotoxic response against nodavirus-infected cells showed significantly increased GzmB activity whilst this pattern failed to do so in sea bass leucocytes. Moreover, flow cytometry studies also suggested the presence of GzmB into the cytoplasm of effector cells. At gene level, seabream GzmB gene was found to be up-regulated in leucocytes under cytotoxic activity, mainly if the target cells were infected with nodavirus. These data will through some light in the importance of granzymes in the cytotoxic response of fish and their defense against nodavirus infection.

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